

Hesperapis rhodocerata: Behavioral Biology, Egg, and Larval Instars, Including Behavioral and Larval Comparisons with *H. larreae* (Hymenoptera: Melittidae: Dasypodainae)

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ABSTRACT

This paper reports on a large nesting site of the ground-nesting solitary bee *Hesperapis* (*Carinapis*) *rhodocerata* (Cockerell) from southern New Mexico first discovered in the late summer of 2010 and active again in late summer 2015. Because the site was visited annually during intervening years without observation of any specimens, the species is believed to sustain a multiyear diapause that is broken in response to rain. It is judged to be univoltine, and females at the site collect pollen from *Heterotheca* (Asteraceae). Nests are briefly described as are the nest-digging behavior and pollen-transport system of females. The feeding behavior of larvae involves grazing on the surface of the food sphere, thus reducing its diameter. This is accomplished with the aid of paired ventral tubercles on each of the three thoracic and first eight abdominal segments and a single median ventral tubercle on the ninth abdominal segment. The second and last larval instars are described and illustrated. The first instar is essentially identical to the second instar except for size. Mature larvae are similar to other known *Hesperapis* larvae. The strongly curved egg of *H. rhodocerata* is described and illustrated with a diagram and SEM micrographs of the micropyle.

Because the last larval instar does not spin a cocoon and freshly constructed brood cells are unlined by females, questions are evoked concerning humidity control and parasite exclusion during the long diapause of mature larvae. This information is compared with and found

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in some ways different from that uncovered in an earlier study of *H. (Amblyapis) larreae* Cockerell. It is hypothesized that the clear thin transparent material covering the postdefecating larva of *H. rhodocerata* may function to inhibit desiccation and furthermore may be the same material that hardens and waterproofs the cell walls of other congeneric species including *H. larreae*, thereby serving a similar function but in a different way.

Because too few mature larvae of *H. larreae* had been collected at the time of drafting the study of that species, their description is added here as an addendum.

INTRODUCTION

Adults of *Hesperapis (Carinapis) rhodocerata* (Cockerell) are moderate in size compared with other *Hesperapis* species and are known from a number of localities in southeastern Arizona, southwestern New Mexico, and adjoining areas in northern Mexico (Ascher and Pickering, 2015). The first nesting site of this species was discovered at 2 mi southeast of Willcox, Cochise Co., AZ, where the nest-digging behavior of females was described, although nests themselves were not explored (Rozen, 1987). The second nesting site was found on August 28, 2010, in a remote area containing few human habitations at 28 mi south of Animas, Hidalgo Co., NM. It was discovered by the author, John S. Ascher, and other attendees of Bee Course 2010. Female bees were digging tunnels in a broad area, perhaps 75 m in diameter on slightly sloping ground just west of low hills (fig. 1). The ground surface was extensively covered with grass, low growing herbs, and scattered yucca plants. In addition, the area supported a large amount of *Heterotheca* (Asteraceae), the pollen source of the large nesting population of *H. rhodocerata*. Intriguing was the observation of numerous individuals of a rather large species of the cleptoparasite *Sphecodes* (Halictidae), whose body size seemed to match that of *H. rhodocerata*. Could this be a nest parasite of *H. rhodocerata*?

In an attempt to explore this question we excavated several nests the same day (fig. 2) and learned that the nests were difficult to follow because tunnels twisted and turned and cells were difficult to associate with their nest tunnels. Although we found no evidence of *Sphecodes* in cells, we did recover two eggs of *H. rhodocerata* (described below) and some early larval instars of *H. rhodocerata*. Because the nesting site was extensive and densely populated, it would likely persist and therefore suggested that it could be explored further at about the same time the following year.

After the end of the Bee Course the collection dates of specimens of this species were checked in the American Museum of Natural History (AMNH) and in Ascher and Pickering (2015). Adults have been collected only during August and September. This indicates that *H. rhodocerata* is a late summer, univoltine species (later also confirmed by J. Neff, personal commun., X-25-2015). Although this observation provided more promise that this large population would reappear in 2011, it failed to do so. The site was checked about the same time every year thereafter until 2015. Although checking the site usually involved only a single annual visit, it never resulted in the sighting or capture of any specimens much less in the appearance of a massive emergence. However, in 2015, following extensive summer precipitation in the area, the large populations of *H. rhodocerata* and *Sphecodes* reappeared in the presence of extensive



FIGURES 1– 4. Nests of *Hesperapis rhodocерata*. 1. Nesting site at 28 mi south of Animas, Hidalgo Co., NM, with most nest entrances among yellow-flowered *Heterotheca* toward right side of picture when first found in late summer 2010. 2. John S. Ascher excavating first nest in 2010, with Margaret A. Rozen looking on. 3. Female *Hesperapis rhodocерata* with pollen loads being transported on anterior surface of hind tibiae. 4. Close-up of basitarsus showing dorsal trough of hairs used for flinging sand. 5–9. Sequential stills from slow-motion video showing position of hind legs when flinging sand from nest entrance. For explanation, please read text. [Figs. 1–3 courtesy John S. Ascher]

flowering of *Heterotheca*. This large emergence, with no change in the site throughout the intervening years, strongly suggests that the 2010 population was the immediate parent of the 2015 population. The following is a report of what was learned about the nesting biology of *H. rhodocrata* and comparative descriptions of the eggs discovered in 2010 and larvae from 2010 and 2015. Field observations in 2015 occurred from August 20 through September 13. Specimen preparation and examination continues even while this manuscript is prepared for press.

In several parts of this paper, I refer to a work entitled “North American Melittidae (Doc. 143153),” comprising pages 14–96 of typewritten manuscript by the late Gerald I. Stage.² It pertains to the biology of the genus *Hesperapis* and the larger manuscript ends with a series of tables (pp. 406–410). Stage presented it to me years ago before the publication of Rozen and McGinley (1991), therein referred to as “Stage, G.I., and R.R. Snelling, unpublished ms. A revision of Nearctic Melittidae: the subfamily Dasypodinae (Hymenoptera: Apoidea).” It was edited by him with Snelling’s name written in ink by Stage, suggesting that it would have been published jointly by them (Stage and Snelling, ms). It covers the following subtopics: life history, nest note, mating behavior, nest construction, nest architecture, provisions, foraging behavior, and sleeping behavior with respect to seven species, two of which presumably have yet to be described.

METHODS

For study, all preserved larvae were cleared in an aqueous solution of sodium hydroxide after heads were removed from bodies, stained with Chlorazol Black E, and then examined and stored in glycerin on well-slides. The egg was critical-point dried and coated with gold/palladium before being examined with an Hitachi S5700 scanning electron microscope.

Scale bars on all diagrams = 1.0 mm.

NESTING BEHAVIOR

Nest openings were scattered throughout the area mostly on flat surfaces between plants, usually surrounded by loose, course soil. Because a large species of *Agapostemon* was also nesting throughout the area, it was impossible to differentiate between nests of the two species unless a female was observed entering or cells were uncovered holding identifiable immatures.

NEST STRUCTURE AND CONFIGURATION: To best understand the configuration of the nest of a ground-nesting bee it is desirable to select a single nest well separated from other nests and to carefully dissect the soil from one side of the descending main tunnel, as detailed in Ramos and Rozen (2014) for *Psaenythisca wagneri* (Vachal). Thus, confusion with neighboring nests is avoided. However, the area selecting for excavation was chosen because the abundance

² Stage was obviously a keen observer and an articulate writer. Anyone seriously studying the biology of this genus should contact me and request a copy of the part of the manuscript available.

of nest entrances promised recovery of numerous cells of *H. rhodocерata*. Consequently, information concerning the nest is limited. It consisted of branching, mostly open tunnels 5 mm in diameter, twisting and turning as they extended downward. Ovoid cells about 15 mm long and 10 mm in maximum diameter were arranged singly and approximately horizontally, mostly between depths of 21–36 cm below the surface. Their walls were rough, without any evidence of a special lining. In general nests were therefore similar to that diagrammed for *H. (Hesperapis) trochanterata* Snelling (Rozen, 1987: fig. 3).

NEST-DIGGING BEHAVIOR: As first reported by Rozen (1987), nest excavation by females of *Hesperapis rhodocерata* and *H. trochanterata* near Willcox, Cochise Co., AZ, involves an extremely rapid flinging of surface sand backward under their bodies at nest entrances. This behavior also occurs when a foraging female returns to her nest entrance. The result is a tumulus of fine loose sand that widely surrounds the entrance except at the very center from where the female discharges the soil. This was photographed for *H. trochanterata* (Rozen, 1987: fig. 2) where the sand was dry, uniformly fine, so that the tumulus formed a smooth mound. At that time it was determined that the female's forelegs dug sand from the entrance and the hind legs flung the excavated sand backward so rapidly that the exact motion of the hind legs blurred.

Now with the advent of smartphones (e.g., iPhone 6sTM) with slow-motion video capability, a more detailed analysis was forthcoming for *H. rhodocерata* at 28 mi south of Animas, Hidalgo Co., NM (taped on IX-28-2015). To interpret the actions of the female in the video, one must be aware of the anatomy of the female's hind leg. The leg bears unusually long scopal setae on the anterior surface of the tibia and lacks any such setae on the femur. The dorsal surface of the basitarsus (fig. 4) has a long, narrow, barren strip sharply defined for almost its entire length by two parallel rows of long, slightly curved setae forming the sides of a trough extending nearly the length of the basitarsus. The anterior surface of the basitarsus bears a continuation of the scopal plumage, while the posterior surface continues the short vestiture of the posterior tibial surface.

In the video, the returning female *H. rhodocерata* had both scopae filled with pollen, and the nest entrance was clogged with a mixture of sand and gravel obstructing her reentry. With her head partly inserted into the entrance tunnel, she rapidly moved her forelegs, thereby raking the sandy mixture from the hole toward the ground surface under her body (fig. 5). Her mid legs, partly flexed, extended laterally forming an anchor, so that she maintained her position relative to the entrance hole. The bracing is required to establish a stable platform to counteract the rapid action of her front legs and the strong strokes of her hind legs. Without slow motion the strokes were apparent because they resulted in flying sand and pebbles, but with slow motion, the rhythmic strokes are seen as a repetitive, nearly simultaneous performance by both hind legs. It starts with the tight folding of the femoral-tibial joint on each side of the body (fig. 5), followed by the forceful unfolding of both legs backward along the side of the metasoma (figs. 6, 7) and then outward (figs 7, 8) as she flings sand and pebbles backward and outward primarily from the troughs of both basitarsi. Immediately following this action she swings both legs back (figs. 8, 9) to the starting position (fig. 5) for the next fling. As she

continues this routine, her body direction gradually changes relative to the entrance hole so that the tumulus tends to accumulate on all sides of the hole.

POLLEN TRANSPORT: Although females acquire pollen on many surfaces of their bodies because of long body vestiture, the tibial scopae are the structures on which females accumulate large agglutinated masses of pollen (fig. 3) to be transported to the nest. Because scopal hairs are restricted to the anterior surface of the hind tibia, these masses are found there and do not surround the tibia. Very large tibial masses seem to extend apically over the basal part of the dorsal surface of the basitarsus, but it is uncertain whether the setae there are sufficiently long to hold the mass. However, the anterior row of long setae bordering the dorsal trough appears to fence off the pollen from invading the setal trough of the basitarsus. It is unknown how many provisioning trips are required to form the final ball of provisions, but the total amount is shaped into a sphere that is approximately 5.3 mm in diameter and placed on the cell floor (fig. 11).

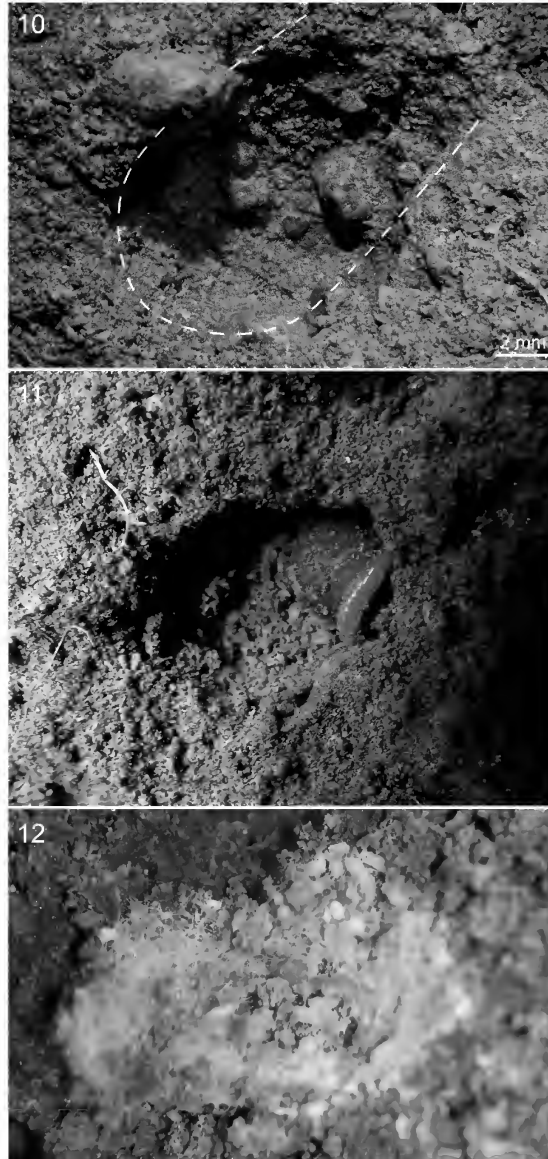
LARVAL BEHAVIOR: Numerous observations indicate that the size of the feeding larva increases as the diameter of the food sphere diminishes. However, the provisions do not change shape, thus indicating that the larva is feeding throughout the surface of the sphere. Masses, fresh as well as partly eaten, randomly collected from nest excavations during the field season in 2015, ranged from 5.3 mm in diameter down to 2.6 mm but always retained their approximate spherical shape. The larval movements seemed unusually slow. The following description, referring to an unnamed species in the unpublished manuscript by Stage, is consistent with my fragmentary observations:

The strongly curved, cylindrical egg is placed on top of the pollen ball in such a way that only its ends are in contact with it.... The duration of the egg stage is not known but presumably is short as in most other bees.

Upon eclosion the small larva starts feeding near the top of the pollen ball.... While still small it gradually works its way down and around the pollen ball until it achieves a characteristic position curled around and under the pollen ball.... At this time the larva is in a C-shape with one of its sides, not its dorsum, against the floor of the cell and its venter against the pollen ball. In this position the larva continues to feed but at the same time it slowly rotates the pollen ball by a constant twitching motion of the terminal segments of the abdomen. As the larva becomes larger this action tends to lift the pollen ball so that it becomes entirely supported by the larva. The effect of this unusual feeding behavior is that the pollen ball is evenly grazed and remains nearly spherical until it has been almost completely consumed. (Stage and Snelling, ms: 18)

The anatomy of early instars, described below with paired ventral body tubercles and spiculate ventral integument, would seem to be suitable for crawling over the surface of the sphere and then for lifting it up and rotating it while feeding.

NEST CELL ENVIRONMENT: A series of recent studies points out that bee cocoons function to maintain appropriate humidity around the diapausing larva and developing pupa while



FIGURES 10–12. Macrophotograph of cells of *Hesperia rhodocerata*. **10.** Empty cell, dorsal view showing coarse surface similar in texture to that of surrounding substrate. **11.** Cell, top removed, showing second or third larval instar on top of sphere of provisions and rough, uneven surface of cell floor without special lining. **12.** Fragment of cell floor covered with moldy fecal pellets. [Fig. 11 courtesy William de Oliveira Sabino]

preventing attacks by nest parasites and predators (e.g., Rozen and Hall, 2011). It is likely that special cell linings constructed by nest-making females form a partially waterproof barrier and also contribute to humidity control. Larvae of *H. rhodocerata* as well as all other *Hesperapis* (and presumably those of *Capicola* and *Dasypoda*) do not spin cocoons (Rozen, 1974; Rozen and McGinley, 1974). Furthermore, females of many (though presumably not

all³) *Hesperapis* whose nests have been examined do not provide a specially prepared, water-retardant lining to their brood cells (Rozen, 1987; Stage and Snelling, ms.). How is the diapausing larva protected from desiccation between flowering seasons of the host plant? Perhaps the following observations hint at an explanation.

A last larval instar was preserved as a postdefecating form on IX-15-2015. When brushed three weeks later to remove sand grains prior to being illustrated, its body surface was found to be completely covered by a thin transparent coating, which floated away from the surface as thin, nearly transparent flakes in the preservative. This solid material had not been detected until the brushing. Might it have to do with maintaining body moisture? On another specimen, this material was carefully removed from around several spiracles. While maintaining the surface feature surrounding the spiracular opening, the opening itself was not covered, i.e., it remained an aperture (fig. 35). Almost identical observations had been recorded regarding *H. trochanterata* (Rozen, 1987), although then the material was described as “tannish.” One wonders if this coating helps to retain body moisture during the long larval diapause. With both species it is evident only on the postdefecating larva. Still unknown is the source of this coating and chemical nature of the substance.

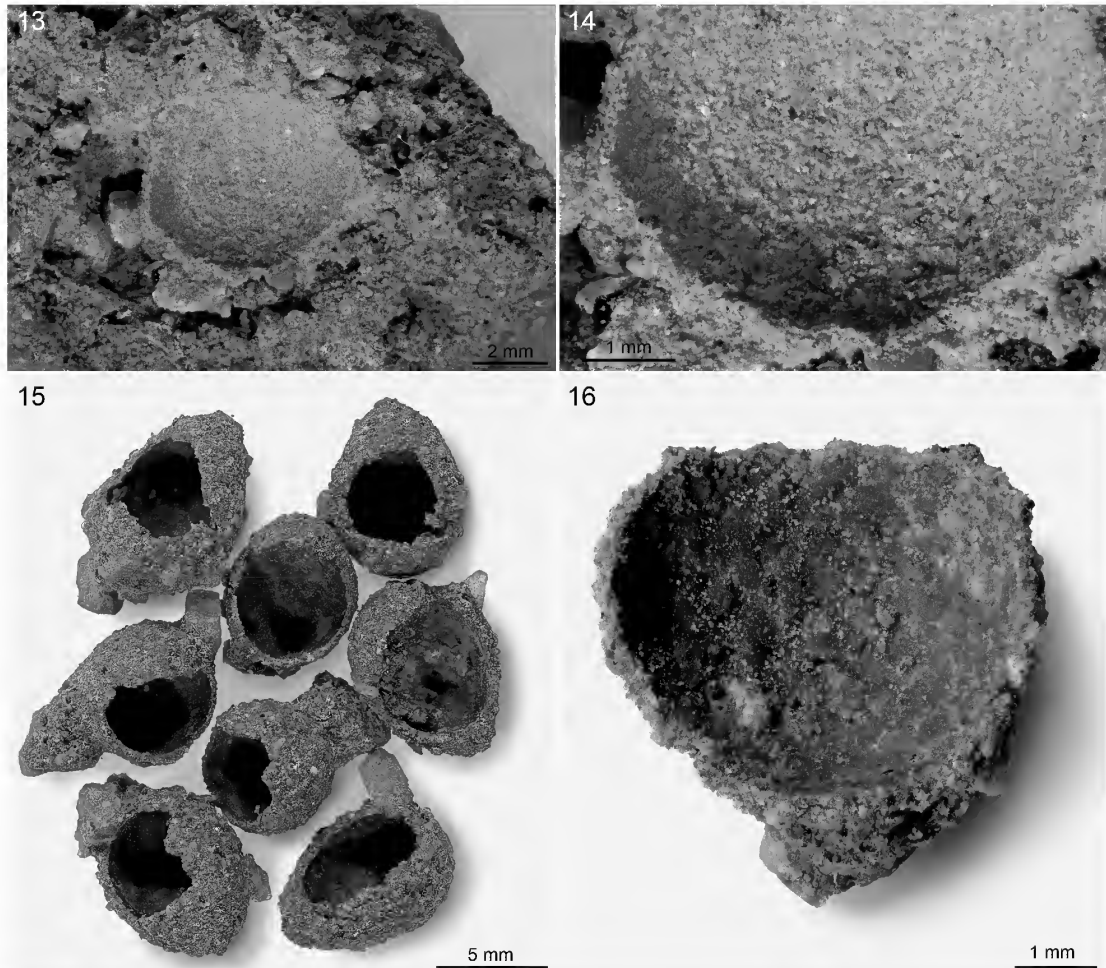
A more recent study pertaining to the behavioral biology of *H. (Amblyapis) larreae* Cockerell (Rozen and McGinley, 1991: 5–6) presented a somewhat different story. It stated that recently constructed cell walls of this species

tended to be slightly more consolidated than the substrate. Small sections of wall could often be carefully teased from the substrate.... Either after feeding or perhaps shortly before finishing, large larvae produced a substance (source unknown but perhaps anal or salivary) that impregnated the cell wall and closure so that these structures became strong, took on a dark ‘wet’ appearance, glistened in places, and became water-retardant.⁴ This substance possessed no pollen grains (at least at first) and a section of impregnated cell wall did not ‘dissolve’ when submerged in water for several hours. Because of their new strength, cell walls and closures in this condition were extricated intact from the substrate.

In the same article: “Fresh walls were not waterproof and immediately absorbed water droplets. They were smooth, dull on the surface, and gave no hint as to what substance (if any) accounted for their slightly greater strength than the substrate. The soil of the cell wall was uniformly fine-grained in sharp contrast to the irregular particle size of the surrounding substrate. *Hesperapis* females are apparently capable of sorting out fine particles to construct the

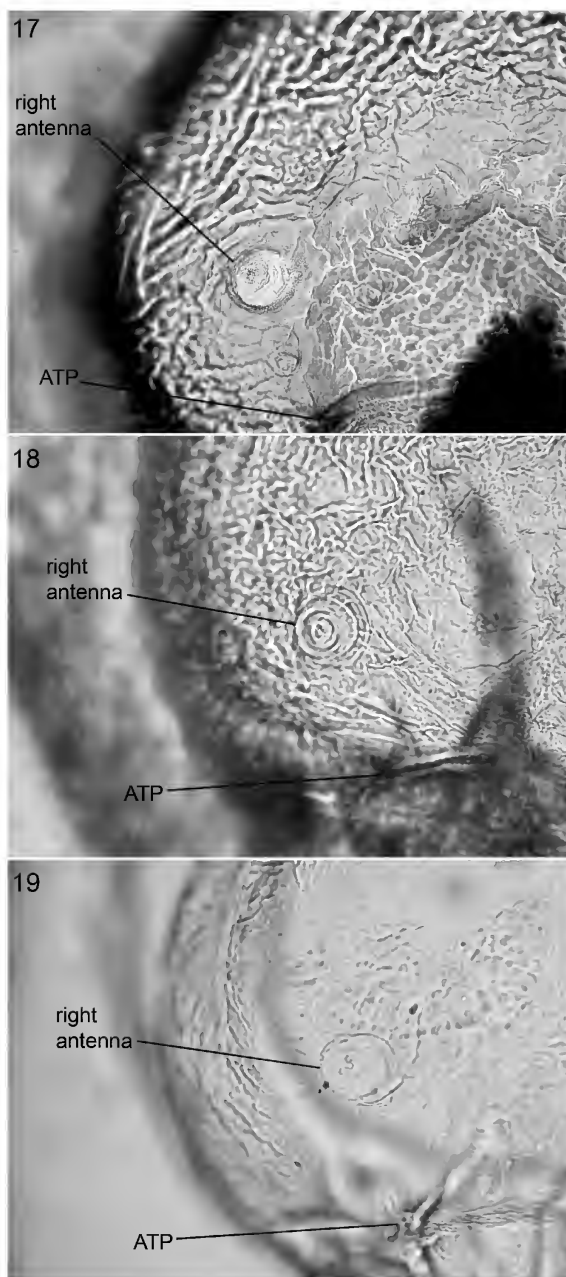
³ Stage and Snelling, (ms: 70) reported that *H. (Hesperapis) rufipes* (Ashmead) and *H. (Amblyapis) ilicifoliae* (Cockerell) each had a cell “with a hard, smooth wall that was constructed on the crude surface of the coarse, heterogeneous substrate and that was apparently smoothed with the tongue and hardened with saliva.” However, there is no indication in their report that cell walls at least of some *Hesperapis* are modified by the mature larva (Rozen and McGinley (1991).

⁴ A small piece of the cell wall collected in 1990 and preserved in the AMNH was placed in water for three days as this paper was being drafted. The piece remained unchanged, attesting to the stability of its waterproof condition.



FIGURES 13–16. Macrophotographs of cells of *Hesperapis larreae* collected in 1990 and 1994, preserved in the AMNH, from extreme southern Yavapai Co., AZ, at 8 mi NW of Wickenburg, Maricopa Co. Note smooth inner surface of wall and fine-grained sand lining contrasting with coarse substrate. **13.** Fresh cell. **14.** Close-up of cell wall, showing fine texture and absence of protruding pebbles. **15.** Cells from which postdefecating larvae removed after application of hardening material. **16.** Fragment of cell wall from figure 15 showing brownish, smoothly flattened fecal pellets.

thin walls” Rozen and McGinley, 1991: 6). Although no photographs of the cells were taken at the time, figures 13 and 14 are photographs of cells of *H. larreae* collected a few years later and preserved in the AMNH. The sharp contrast of the small particle size of the wall with the irregular, often coarse particle size of the substrate is undeniable. Note that the surface of the cell wall is without larger pebbles invading the cell lumen in contrast to the surface of the cell wall of *H. rhodocerta* (figs. 10, 11). Figures 13 and 14 show the wall of cells containing provisions are distinctly different from the substrate surrounding them even though they have not yet been hardened by the mature larva. Figure 15 shows the distinctly hardened cell walls and cell closures that had contained postdefecating larvae.



FIGURES 17–19. Microphotographs of right side of cleared head capsules of three species of *Hesperapis* used to compare degree of integumental wrinkling, frontal view. (ATP = anterior tentorial pit.) 17. *H. rhodocera*. 18. *H. trochanterata*. 19. *H. larreae*.

Although Rozen and McGinley (1991: 6) noted “One larva had started to apply fecal pellets over the inner surface of the cell wall...,” their observations were too early in the life of the larva to comment further. Completed cells of this species, collected in 1994, reveal brown, smoothly flattened pellets presumably concentrated on one part of the cell surface, as shown in figures 16. This contrasts with the appearance of fecal deposited in cells of *H. rhodocерata* (fig. 12). These deposits, though scattered, appeared densest on the cell floor, and are rough projecting pellets, supporting dense white mold, all of which may be the result of having only a rough, uncoated cell wall.

The following behavioral similarities are shared by *H. rhodocерata* and *H. larreae*: (1) the cell walls are absorbent to water droplets until the larvae are fully grown; (2) the substance (or substances) produced by the larvae before entering diapause either coats their bodies (*H. rhodocерata*) or hardens the cell walls and closures (*H. larreae*), depending on the species. Where do these materials come from and are they the same? How does the material spread over *H. rhodocерata*? The larva of *H. larreae* does not have strong surface wrinkles (see Addendum, below, in which the mature larva of *H. larreae* is described). Stage and Snelling (ms) stated that the inner surface of the cell wall is smooth for both *H. (H.) rufipes* and *H. (Amblyapis) ilicifoliae*. Mature larvae of these two species (from Stage collection) are not strongly wrinkled (or “less tough and ridged,” sensu Stage and Snelling, ms: 22), although the larva of the former is somewhat more wrinkled. The larva of *H. larreae* is known to harden the cell wall and closure with some liquid that wets the wall and later renders it waterproof. In contrast, the larva of *H. rhodocерata* has a wrinkled integument and seemingly produced a material that covers the integument. These facts suggest the following hypothesis: the material that both larvae produce may be the same and in *H. rhodocерata*, it is distributed by spreading over the larva’s integument by capillarity whereas, in *H. larreae* it is absorbed by the sandy lining of the cell and accounts for the hard waterproof surface of the lining, which develops thereafter.

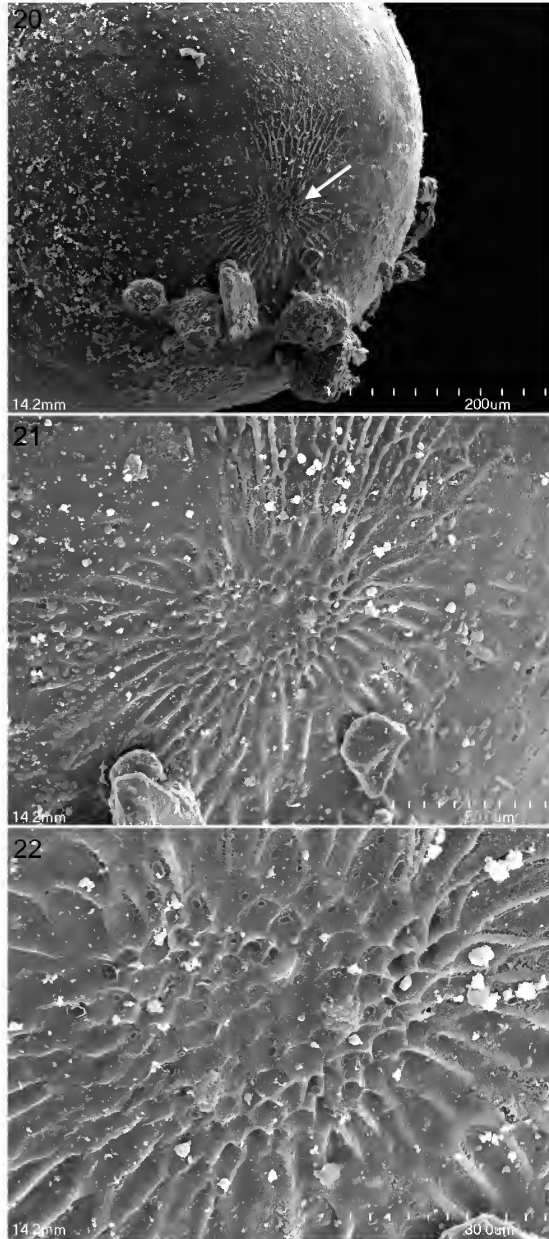
DESCRIPTION OF IMMATURE STAGES

EGG

Figures 20–23

These specimens were collected and preserved when the site was first discovered. The anterior end was determined by the orientation of the embryos. The larger of the two eggs seemed to be better preserved and therefore was used for illustration (fig. 23). Length 2.7, 2.95 mm; maximum diameter (front end) 0.58, 0.75 mm. Color white; shape strongly curved; front end rounded; posterior end smaller but rounded; chorion smooth, transparent. Under SEM chorion without sculpturing except at anterior end, ridges radiating around micropyle that consists of a cluster of pores (figs. 20–22).

MATERIAL STUDIED: Two eggs: NM: Hidalgo Co.: 28 mi S. of Animas, August 30, 2010 (J.G. Rozen).



FIGURES 20–22. SEM micrographs of front end of egg of *Hesperapis rhodocera*. 20. Showing position of micropyle (arrow). 21. Close-up of same, showing micropyle and surrounding sculpturing of chorion. 22. Close-up of micropyle.

SECOND LARVAL INSTAR

Figures 24, 25

The specimens described here are believed to be the second instar based upon the assumption that this bee has five larval instars, which seems likely based on a survey of head sizes of collected specimens. The second instars were selected from among the available specimens of earlier instars because they best illustrated the distinctive, unusual features of earlier larval instars of *H. rhodocерata* due to the quality of their preservation and size compared with both smaller and larger specimens. The following description indicates how the anatomy of this instar differs from that of the last larval instar.

DESCRIPTION: Body shape extremely elongate, with dorsal and ventral surfaces nearly parallel in lateral view (fig. 24), so that body not tapering posteriorly as in mature larvae (figs. 26, 27); body segments with cephalic and caudal annulets not defined; thoracic segment and abdominal segment 1–8 each with pair of ventrolateral tubercles (prolegs); abdominal segment 9 strongly produced ventrally on first half of segment. Ventral integument including that of tubercles and ventral surface of abdominal segment 9 uniformly, extensively, but finely spiculate.

MATERIAL STUDIED: One second larval instar: NM: Hidalgo Co.: 28 mi S. of Animas, September 6, 2015 (J.G. Rozen, B. Lawley). One second larval instar: same, except August 30, 2010 (J.G. Rozen, J.S. Ascher).

REMARKS: These specimens were compared with a first instar collected at the same locality on October 30, 2010, by J.G. Rozen and J.S. Ascher. Although smaller than the second instar, the first instar otherwise agreed completely with the above description.

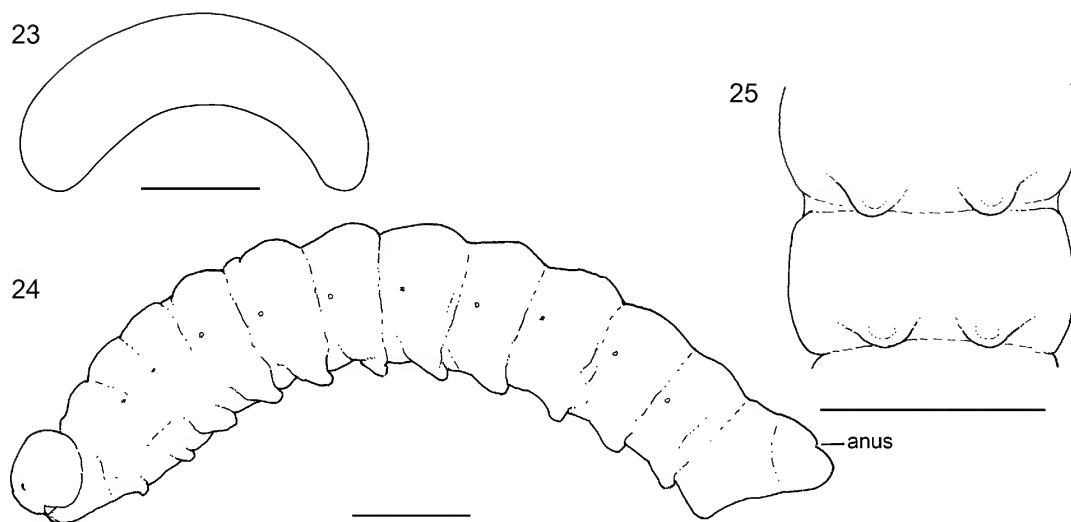
The third instar, while retaining the linear appearance and spiculated venter, has the ventrolateral tubercles less pronounced. In the fourth instar spicules persist but paired ventrolateral tubercles have virtually disappeared, although the basal swelling of the venter of abdominal segment 9 is retained, so that in lateral view, that segment is as wide as the preceding one. In the last larval instar described below, there is no hint of paired ventrolateral tubercles.

MATURE LARVA

Figures 26–29, 31–34

DIAGNOSIS: The mature larva of *H. rhodocерata* agrees closely with described larvae of other species in the genus (Michener, 1953⁵; Rozen and McGinley, 1974; Rozen, 1987, and references therein) in sharing in lateral view an elongate body form with caudal annulets of body segments scarcely projecting farther than cephalic annulets, abdominal segment 10

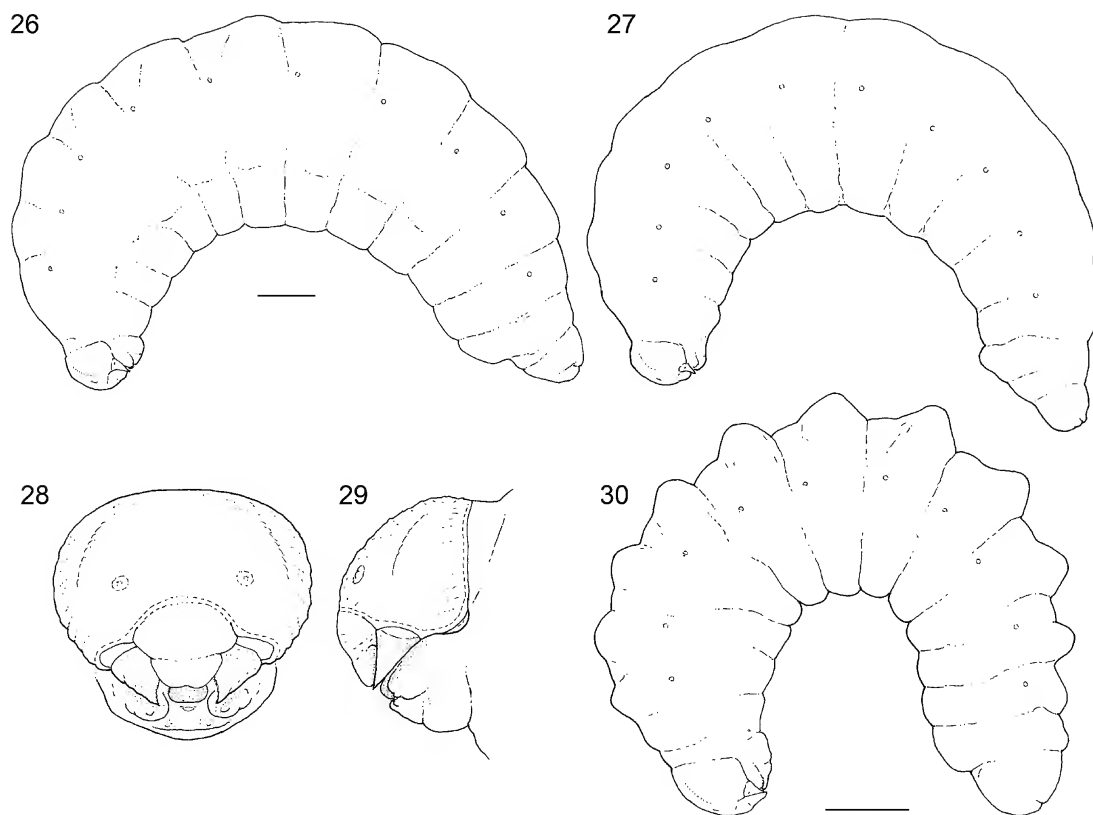
⁵ Michener (1953) provided the first description of any larval *Hesperapis*, i.e., *H. rufipes* (Ashmead). Although he stated that the epistomal suture was not recognizable, the internal epistomal ridge is visible before fading out toward the head midline.



FIGURES 23–25. Diagram of egg of *Hesperapis rhodocerata* lateral view, anterior end to left. FIGURES 24, 25. Diagrams of second instar of *Hesperapis rhodocerata*, spicules not shown. **24.** Entire larva, lateral view. **25.** Abdominal segments 4 and 5, ventral view showing paired tubercles.

attached dorsally to 9th segment; and venter of abdominal segment 9 much longer than the dorsum of same segment. Furthermore, the cranium of known species is much broader than its height in frontal view. In lateral view the profile of the head from vertex to labral apex forms a continuous even curve (fig. 29).

DESCRIPTION: Head: Integument of head capsule and labiomaxillary region with surface extremely wrinkled but substantially more deeply so on postdefecating form than on predefecating form; pigmentation evident primarily on mandibular apex and teeth, but integument of head capsule tending to be slightly darker than that of body presumably because of thickness; long setae entirely absent but minute setiform sensilla present; extensive spiculation on dorsal surface of maxilla and hypopharynx. Cranium wide in frontal view, with width distinctly greater than distance measured from lower clypeal margin to top of vertex. In lateral view profile curving forward, so that frontoclypeal surface farthest forward; antenna prominence not present. Tentorium complete, including dorsal arms and posterior bridge. Internal head ridges well developed except epistomal ridge becoming obscure near midline (fig. 28); as in most if not all other known *Hesperapis* and *Capicola capicola* (Fries), dorsal ramus of hypostomal ridge well developed and curving to join posterior thickening of head capsules (see remarks, below); longitudinal thickening of head capsule absent. Antenna not on elevation; antennal papilla very short, disclike, bearing sensilla. Parietal bands sharply defined. Labrum moderate in size, with apex not projecting nor bearing lateral swelling or tubercles; apical labral margin straight to faintly bilobed, curving toward clypeal margin on each side. Mandible, as seen in inner or outer views, tapering evenly to sharp apex; apical concavity only faintly evident and without teeth along ventral edge; surface of concavity filled with numerous ridges leading to teeth along dorsal edge and without spicules; dorsal inner edges covered by narrow band of sharp tubercles; base of entire mandible covered



FIGURES 26–30. Diagrams of mature larvae of *Hesperapis rhodocерata*. **26.** Entire postdefecating larva, lateral view. **27.** Predefecating larva, lateral view. **28, 29.** Head, frontal and lateral views, respectively. **FIGURE 30.** Diagram of mature larva of *Hesperapis larreae*, lateral view.

with spicules. Labiomaxillary region recessed in lateral view; lobe of hypopharynx in lateral view extending slightly beyond maxillary apex, which extends only slightly beyond labial apex in lateral view; maxillary palpus short, moderately stout; maxillary sclerites and galea not expressed. Hypopharyngeal groove well developed; articulating arm of stipes not visible. Labium weakly divided into pre- and postmentum, salivary opening simple hole without lips; labial palpus faintly projecting, evident mostly because of sensilla.

Postcephalic region: Integument of postdefecating form stiff, at least in part because of pronounced fine wrinkling, compared with that of predefecating form, which is pliable and far less wrinkled; spiculation on postdefecating form detected only after faint ventral spiculation found on predefecating form. Body form of postdefecating larva moderately robust, tapering posteriorly in lateral view; intersegmental lines moderately incised; paired dorsal tubercles absent; dorsal intrasegmental lines weak; caudal annulets scarcely defined; abdominal segment 9 with venter elongate, projecting ventrally at base, then narrowing abruptly in lateral view, so that segment 10 appearing to attach to it dorsally (figs. 26, 27); anus somewhat dorsal on abdominal segment 10; perianal area strongly wrinkled. Spiracles (figs. 33, 34) moderately

large, subequal in size, pigmented; atrium with projecting rim, nearly spherical in shape with width about same as length, and with diameter of rim (fig. 34) distinctly smaller than diameter of atrium; atrial wall without ridges or spicules; primary tracheal opening with strong collar; subatrium with outer chambers distinct, moderately sclerotized, inner chambers less sclerotized, often collapsing one against another, hence length difficult to judge.

MATERIAL STUDIED: Four postdefecating larvae: NM: Hidalgo Co.: 28 mi S. of Animas, September 8, 2015, preserved September 15, 2015 (J.G. Rozen, B. Lawley). Five predefecating larvae, same locality and collectors, collected and preserved September 8, 2015.

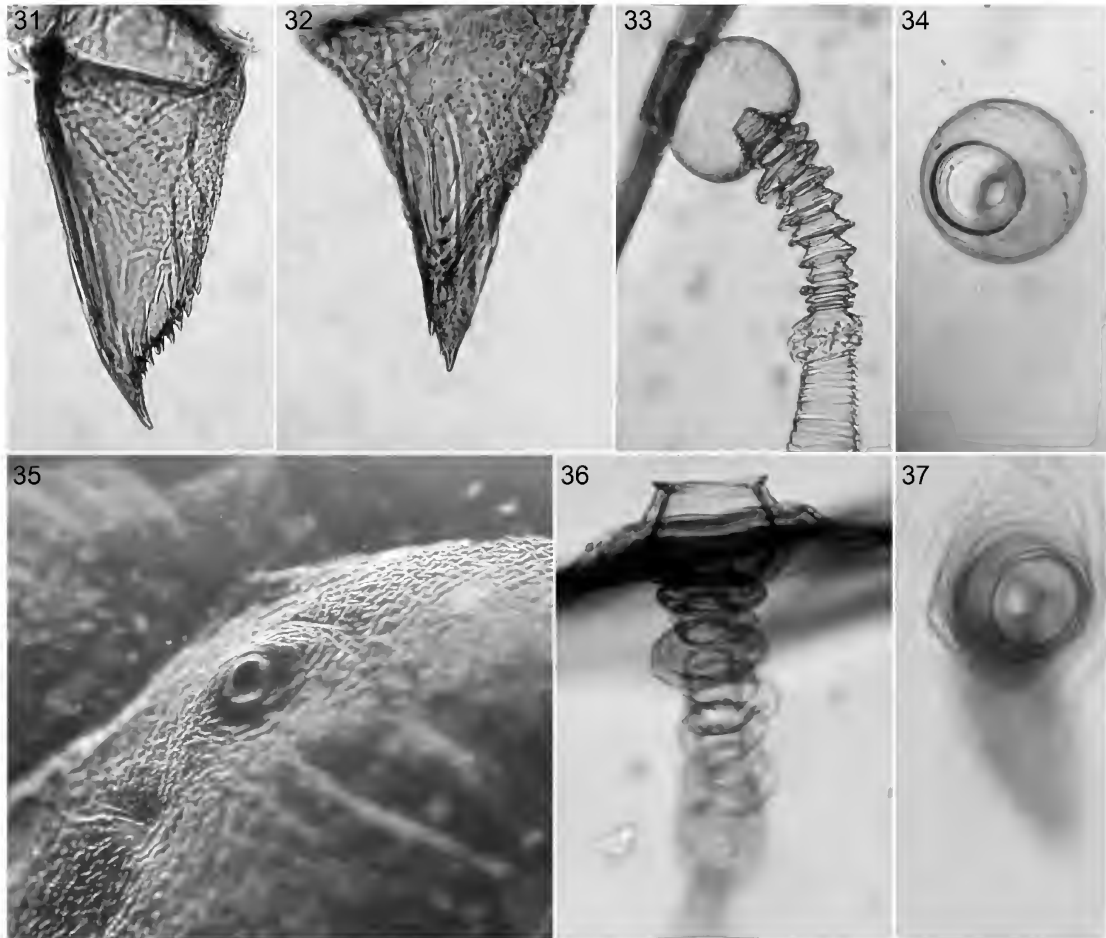
REMARKS: Rozen and McGinley (1974) were able to distinguish the sex of at least some of the species of mature larvae of *Hesperapis* involved with their investigation. A feature for recognizing females involved identifying imaginal disc responsible for adult female structures seen through the predefecating larva's ventral integument. This sex-recognition method did not reveal any when applied to *H. rhodocerata*, possibly because the imaginal discs were not revealed against the white background of the predefecating form. However, one cleared and stained postdefecating larva bore cuticular scars on the venter of abdominal segments 7–9, visible because of the stain, which was not employed at the time of the earlier study. This suggests that with examination of more specimens, identification of the sex of mature larvae will become reliable.

In early treatments of melittid larvae, the dorsal ramus of the hypostomal ridge was not understood. It is now recognized as a secondary strengthening structure that extends posteriorly from about the middle of the hypostomal ridge at the point where the hypostomal ridge bends sharply mesad to connect to the posterior tentorial bridge at the posterior tentorial pit. In Rozen and McGinley (1974: fig. 64) the small triangular part of the parietal encompassed by the hypostomal ridge, the posterior thickening of the capsule, and the dorsal ramus was referred to as "swelling." The ramus is absent from known larvae of *Meganomia*, *Melitta*, *Macrotera*, and seemingly incomplete in *Dasypoda* (Rozen, 1977, 1978; Rozen and Jacobson, 1980).

DISCUSSION

This study confirms the uniformity of mature larval anatomy within the genus *Hesperapis*, characterized by a broad head capsule that is almost hemispherical in lateral view and an elongate body ending with a narrow abdominal segment 10 attached dorsally to segment 9. These features are shared with *Capicola capicola* and less so with *Dasypoda plumipes* Panzer (Rozen and McGinley, 1974).

The biological information here highlights an interesting question: how can the larva of such a small bee survive while in diapause over a long to very long period when it must remain dormant until environmental conditions permit production of its food plant? It has been demonstrated that many bees (all Megachilidae, many Apidae) have cocoons that regulate cell environments, others (most Colletidae, many if not all Andrenidae) have cells in which the nest-making females have applied water-retardant cell linings. However, *Hesperapis* larvae lack such protections. Here it is hypothesized that the mature larva of *H. rhodocerata* produces a substance that covers its body with a thin layer of material that reinforces the water-retention quality of its



FIGURES 31–37. Microphotographs of mature larva of *Hesperapis rhodocerata*. 31, 32. Right mandible, dorsal and inner views, respectively. 33, 34. Spiracle, side view and outer views, respectively. 35. Surface covering material from around spiracle. FIGURES 36, 37. Microphotographs of spiracle of mature larva of *Hesperapis larreae*, side and outer views, respectively.

integument. Further, it is pointed out that the mature larva of *H. larreae* produces a liquid that hardens both the cell wall and cell closure making them waterproof, thereby providing the cell inhabitant protection against desiccation and parasite attack. Questions arise: where are the two substances produced, how are they applied, and might the two substances actually be the same material? The next step in this study will require analysis and comparisons of the materials produced by the two larvae and the discovery of their source or sources.

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REFERENCES

- Ascher, J.S., and J. Pickering. 2015. Discover Life. Online resource (<http://www.discoverlife.org/mp/20q?search=Apoidea>).
- Michener, C.D. 1953. Comparative morphology and systematics studies of bee larvae with a key to the families of hymenopterous larvae. University of Kansas Science Bulletin 35: 987–1102.
- Ramos, K.S., and J.G. Rozen, Jr. 2014. *Psaenythisca*, a new genus of bees from South America (Apoidea: Andrenidae: Protandrenini) with a description of the nesting biology and immature stages of one species. American Museum Novitates 3800: 1–32.
- Rozen, J.G., Jr. 1974. The biology of two African melittid bees (Hymenoptera, Apoidea). Journal of the New York Entomological Society 82: 6–13.
- Rozen, J.G., Jr. 1977. Biology and immature stages of the bee genus *Meganomia* (Hymenoptera, Melittidae). American Museum Novitates 2630: 1–14.
- Rozen, J.G., Jr. 1978. The relationships of the bee subfamily Ctenoplectrinae as revealed by its biology and mature larva (Apoidea: Melittidae). Journal of the Kansas Entomological Society 51: 637–652.
- Rozen, J.G., Jr. 1987. Nesting biology and immature stages of a new species in the bee genus *Hesperapis* (Hymenoptera: Apoidea: Melittidae: Dasypodinae). American Museum Novitates 2887: 1–13.
- Rozen, J.G., Jr., and H.G. Hall. 2011. Nesting and developmental biology of the cleptoparasitic bee *Stelis ater* (Anthidiini) and its host, *Osmia chalybea* (Osmiini) (Hymenoptera: Megachilidae). American Museum Novitates 3707: 1–38.
- Rozen, J.G., Jr., and N.R. Jacobson. 1980. Biology and immature stages of *Macropis nuda*, including comparisons to related bees (Apoidea, Melittidae). American Museum Novitates 2702: 1–11.
- Rozen, J.G., Jr., and R.J. McGinley. 1974. Phylogeny and systematics of Melittidae based on the mature larvae (Insecta, Hymenoptera, Apoidea). American Museum Novitates 2545: 1–31.
- Rozen, J.G., Jr., and R.J. McGinley 1991. Biology and larvae of the cleptoparasitic bee *Townsendiella pulchra* and nesting biology of its host *Hesperapis larreae* (Hymenoptera: Apoidea). American Museum Novitates 3005: 1–11.
- Rozen, J.G., Jr., J.R. Rozen, and H.G. Hall. 2011. Gas diffusion rates through cocoon walls of two bee species (Hymenoptera: Megachilidae). Annals of the Entomological Society of America 104: 1349–1354.
- Stage, G.I., and R.R. Snelling. ms. North American Melittidae (Doc. 143153). Supplementary file to this issue (American Museum Novitates 38TK), available online (<http://dx.doi.org/10.5531/sd.sp.17>).

ADDENDUM

DESCRIPTION OF MATURE LARVA OF *HESPERAPIS* (*AMBLYAPIS*) *LARREAE* COCKERELL

Figures 30, 35, 36

DIAGNOSIS: The mature larva of *Hesperapis larreae* exhibits the main larval features of other known members of the genus including linear body form, dorsally positioned abdominal segment 10 relative to abdominal segment 9, anus dorsally positioned on 10, and broad, hemispherical head lacking antennal elevations. Compared with the mature larva of *H. rhodocерata*, it differs in that body size is smaller and the caudal body annulets project distinctly farther than the cephalic annulets and most are produced sublaterally as low, paired tubercles. Furthermore, the acutely pointed mandible bears sharply pointed teeth along the ventral apical edge.

DESCRIPTION: Head: Integument as described for *H. rhodocерata* except for following: integumental wrinkling much reduced and much shallower; head capsule only faintly darker than rest of body. All other head features as described for *H. rhodocерata*, except for following: labrum apically faintly bilobed; integumental ridges of mandible indistinct or absent, but sharp teeth along both dorsal and ventral edges of concavity; hypopharyngeal groove absent.

Postcephalic region: Integument of ventral surface faintly but uniformly spiculate. Form of postdefecating larva moderately robust, gradually tapering posteriorly in lateral view; intersegmental lines moderately incised; body segment with paired low dorsal tubercles; dorsal intrasegmental lines moderately apparent and caudal annulets defined; abdominal segment 9 with venter projecting ventrally at base, then narrowing abruptly in lateral view, so that segment 10 attaches to it dorsally (fig. 29); anus dorsal on abdominal segment 10; perianal area scarcely wrinkled. Spiracles (figs. 35, 36) moderately small, subequal in size, faintly pigmented; atrium with projecting rim, nearly spherical in shape with width about same as length and with diameter of rim (fig. 36) only slightly smaller than diameter of atrium; atrial wall without ridges or spicules; primary tracheal opening with collar; subatrium with outer chambers distinct, moderately sclerotized and broad compared to width of atrium, inner chambers less sclerotized and narrower, decreasing in width.

MATERIAL STUDIED: One larva: AZ: Yavapai Co.: 8 mi NW Wickenburg (Maricopa Co.). Coll. May 7, 1990, as predefecating larva; preserved May 25, 1990, as quiescent postdefecating larva (J.G. Rozen). Seven mature larvae, same locality and collector, May 3, 1994.

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